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Physiological Characterization of mBSA Antigen Induced Arthritis in the Rat. II. Joint Blood Flow, Glucose Metabolism, and Cell Proliferation

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ABSTRACT. Objective. Based on the hypothesis that blood flow in the inflamed joint is inadequate to maintain aerobic glycolysis, we sought to estimate the correlation between blood flow, glucose metabolism, and cellular proliferation rate in the arthritic joint.

Methods. Experiments were performed on rats with antigen induced arthritis (AIA). Regional blood flows (RBF) were measured with the microsphere technique, glucose metabolism by determination of [^{14}C]2-deoxy-D-glucose (2-DG) uptake, and the proliferative response as the incorporation of [^{3}H]-thymidine.

Results. In periarticular soft tissue of the arthritic knee the only significant change in the weight related RBF was an approximate 70% rise on Day 14 after arthritis onset. The RBF was lowest on Day 3 and the time course for the changes was inversely related to intensity of vascular inflammation. Weight related 2-DG uptake was more elevated than the RBF and peaked on Day 3. [^{3}H]-thymidine incorporation in the soft tissue was only markedly enhanced on Day 3. Neither 2-DG nor [^{3}H]-thymidine uptake was affected by treatment with methotrexate or indomethacin. In epiphyseal bone RBF was reduced on the first day of arthritis, but steadily increased thereafter.

Conclusion. In AIA an intense vascular leakiness negatively affects the synovial blood. There is a marked enhancement of glucose metabolism, but only a minor part of this increase seems to be induced by increased cellular proliferation. (J Rheumatol 1998;25:1778–84)

Key Indexing Words:
ACIDOSIS ARTHRITIS BLOOD FLOW
GLUCOSE METABOLISM PROLIFERATION

Clinical findings have reported that there is a local lactate acidosis in the joints of some patients with rheumatoid arthritis (RA). As lactate levels are reported to correlate to the intensity of the arthritis, this variable could be important for the understanding of the arthritis process[1,2]. The acidosis indicates an imbalance where local blood flow fails to meet the metabolic demand. In a complex interaction, both proliferative and vascular inflammatory responses in the joint can contribute to this imbalance; the ingrowth of pannus exceeds the pace of new formation of blood vessels, causing a fall in capillary density (for references see[3]). There is also widespread obliterative microangiopathy creating underperfused areas in the pannus[4]. Furthermore, increased vascular leakiness and subsequent formation of intraarticular effusions would elevate the joint pressure and thus, in turn, compress the synovial blood vessels[3,5]. This is particularly pronounced during joint motion when the intraarticular pressure is reported to rise to levels exceeding synovial capillary perfusion pressure[6].

As both pannus growth and inflammatory activation of the cells in the joint require energy, metabolic imbalance is further aggravated by increased metabolic demand. In patients with RA this can be measured as increased oxygen consumption[7] and glucose uptake[8].

Ischemia can perpetuate the arthritis in several ways; transient periods can induce the formation of oxygen radicals and reperfusion injury[3,6]. Oxygen radicals are also involved in the activation of the transcription factor nuclear factor kappa B (see[9]). Poor nutrition can induce production of heat shock proteins that can activate immunological mechanisms in the development of arthritis (see[10]). Hypoxia itself can also promote pannus growth both by enhancing cellular proliferation[11] and by providing a selection pressure for cells with diminished apoptotic potential, in particular cells with mutations of the *p53* gene[12]. Interestingly, such cells have been reported to be overexpressed in RA synovial tissue[13]. The relationships between these vascular, proliferative, and metabolic factors are, however, difficult to study in humans and little is known about how they are affected in animal models of arthritis.

In a previous study, we found that in collagen induced arthritis (CIA) in the rat vascular porosity was markedly enhanced in joints, but that the changes in blood flow were

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much smaller and non-persistent[14]; these findings concur with the hypothesis of local ischemia in the joints. We deemed the antigen induced arthritis (AIA) model to be more suitable for studies of joint pathophysiology because (1) it has a higher reproducibility than CIA; and (2) only one knee is inflamed in AIA, hence the other serves as an internal control. As in RA, AIA is characterized by 2 essential features; inflammation and formation of aggressive pannus that degrades cartilage and bone[15-17]. In our adjacent work we studied these phenomena simultaneously and found very marked inflammation measured as increased vascular porosity, and which precedes the pannus growth[18]. According to the above hypothesis, this would lead to reduced blood flow which may be insufficient to meet the metabolic demand and, hence, lead to anaerobic glycolysis. To better understand this process, we tried to quantitate both the changes in blood flow and glucose metabolism in the arthritic joint of AIA rats. Also, in a companion article[18] we measured pannus growth as weight increase of the periarticular soft tissue and the effect of methotrexate (MTX) and indomethacin treatment on this variable. In the present study we extend that investigation by assessing the mechanism of pannus formation by measuring the cellular proliferation rate. Further, we determined how this rate is related to glucose consumption and was affected by pharmacological manipulation.

MATERIALS AND METHODS

The methodology is described in our companion work[18]; briefly, experiments were performed on female Dark Agouti (DA) rats (150 g) obtained from Möllegaards Breeding Centre, Køge, Denmark. The experiments were approved by the Animal Ethics Committee of Lund, Sweden. Immunization was performed by intradermal injection with 1 mg methylated bovine serum albumin (mBSA) dissolved in 50 µl saline and emulsified in 50 µl Freund's complete adjuvant. Ten days later the rats were challenged with an intraarticular injection of 50 µg mBSA (1 µg/µl, dissolved in saline) into one knee. The contralateral knee served as a control and was injected with the same volume of saline. Experiments were performed on Days 1, 3, 7, and 14 after arthritis induction.

Anesthesia, general surgical procedure. The rats were anesthetized with a 1:1 mixture of fluansone/fetanyl (Hypnorm®) and midazolam (Dormicum®), both diluted 1:1 with distilled water before mixing and given 0.25 ml/rat subcutaneously. They were tracheotomized, allowed to breathe freely, and placed on a servo-controlled heating pad to maintain body temperature at about 37°C. The right jugular vein was cannulated with polythene tubing for intravenous (iv) administration. One carotid artery was also cannulated and used for reference blood sampling.

Blood flow measurements. The regional blood flow (RBF) of periarticular tissue and of epiphyseal bone were measured using the labelled microsphere method[19-21]. In brief, radiolabelled microspheres are injected into the left ventricle of the heart. The spheres are distributed with the cardiac output and are trapped in the precapillary sphincters. During injection, an arterial blood sample is taken, and based on the relationship between blood weight and radioactivity in that sample and the radioactivity in the studied tissue, RBF can be calculated. In our experiments, one carotid artery was catheterized with polyethylene tubing, which was connected by a Y-connection to a blood pressure transducer and to a peristaltic pump (Alitea, Stockholm, Sweden) for withdrawing the reference blood sample. To prevent clotting all catheters were flushed with heparin (5000 IE/ml) before the experiment. An injection needle connected to a syringe by polyethylene tubing was introduced into the left ventricle of the heart by direct puncture and used for injection of microspheres. The spheres had a diameter of 15 µm and were labelled with ^{141}Ce and suspended in saline with 0.1% Tween 80. The spheres were injected for 30 s and a reference blood sample was withdrawn during the first 2 min from the start of injection. About 1.0×10^6 spheres were given. After the injection a blood sample was taken from the heart needle for determination of the acid base balance and hence localization of the needle, whereafter the animals were killed by overdose of pentobarbital sodium given iv. The anterior part of the periarticular knee tissue was dissected out as described[18]. The remaining part of the knee was regarded as mainly consisting of epiphyseal bone. Samples were weighed and the radioactivity in them and in the reference blood sample was determined by gamma spectrometry. The RBF was then calculated according to the formula:

$$RBF = Q_r \cdot CPM_t/CPM_r$$

where Q_r = reference blood flow (g/min), CPM_t = radioactivity in the tissue sample, and CPM_r = radioactivity in the reference blood sample. Since validity of the microsphere method is dependent on an even distribution of the spheres, the spheres were vigourously shaken in a vortex mixer immediately before injection. Possible distortion was ruled out by determining that blood flow to the kidneys was symmetrical and plausible.

Autoradiographic analysis of glucose uptake. Glucose metabolism was measured as [^{14}C]2-deoxy-D-glucose (2-DG) uptake. This method was originally developed for measuring glucose metabolism in the brain[22] but has also been used in arthritic joints[8]. In the present experiments 15 µCi of 2-DG (56 Ci/mol) was injected iv and allowed to circulate for 40 min, whereafter the rat was killed and rapidly frozen in −70°C alcohol. The frozen knees were sagittally sectioned according to the Ullberg technique[23]. The [^{14}C] content of the tissues was then determined by densitometric analysis of autoradiographs. The densities of the autoradiograms were calculated in 240 × 230 picture elements (pixels) and expressed as a percentage of the value for the corresponding area in the control knee. Densities were calculated for the periarticular soft tissue (anterior and posterior joint capsule/pannus and suprapatellar bursa) and for the epiphyseal bone. n=2 for each day of study.

Quantification of glucose uptake and thymidine incorporation. [^{14}C]2-DG (15 µCi) and [^{3}H]-thymidine (1 µCi; 84 Ci/mmol) were injected iv simultaneously and allowed to circulate for 50 min, whereafter the animals were killed and rapidly frozen in liquid nitrogen. Arterial blood samples were taken 5 min after injection and immediately before the end of the experiment. After thawing, the soft tissue of the knee was dissected out as described above. To relate uptake levels to those of a rapidly proliferating tissue, a piece of small intestine was taken for comparison. All tissue and blood samples were then stored at about −18°C until analyzed. The samples were thawed and then combusted in a Packard 307 oxidizer equipped with Oximate 80 (Packard, Meriden, CT, USA), a robotic system for sample processing. The combusted samples were automatically dissolved in 15 ml Monophase-S (Packard) for [^{3}H] and 12 ml Permaflour-E (Packard) for [^{14}C]. Radioactivity was measured on Tri Carb Spectrometers (Packard). Quench correction was performed by external standard procedures.

In a separate series the modulating effect of MTX (0.3 mg/kg) and indomethacin (2 mg/kg) on these variables was investigated. The rats were treated with an intraperitoneal injection of either compound on the second day of arthritis and the above variable was remeasured the next day.

Substances. Indomethacin (Confortid®) was purchased from Dumex, (Copenhagen, Denmark); methotrexate from Lederle (Wayne, NJ, USA); mBSA, Freund's complete adjuvant from Sigma Chemical Co. (St. Louis, MO, USA); Dormicum® from Roche (Basel, Switzerland); Hypnorm® from Janssen (Beerse, Belgium); microspheres from DuPont NEN (Wilmington, DE, USA); and [^{3}H]-thymidine and [^{14}C]2-DG from American Radiolabelled Chemicals Inc. (St. Louis, MO, USA).

Data, statistical calculations. All data are, unless stated otherwise, expressed as mean ± SEM and are the difference between the value measured in the control and the arthritic knee in the same rat. Statistical significance was calculated using analysis of variance, followed by the Fisher protected least significant difference or, if only 2 groups were compared, by Student's t test. All calculations were performed on an Apple computer using StatView 4.0 (Berkeley, CA, USA) software.

RESULTS

The intraarticular injection of mBSA induced arthritis in all challenged knees, measured as an increase in knee diameter.

The procedure using direct heart puncture for administration of microspheres sometimes causes cardiovascular disturbances that may affect cardiac output. This would likely affect the RBF in both knees alike. We therefore chose to relate the blood flows in the arthritic knees to those in the contralateral control knee. Values are thus expressed as percentage of the weight related (g/min/g) RBF in the control tissue. This also allowed comparisons between studies of changes in blood flow, glucose, and thymidine uptake and weight gain of the pannus. In the periarticular soft tissue, the RBF was significantly elevated only on Day 14. The time course for the changes showed that the mean blood flow of the arthritic knee was lowest on Day 3 of arthritis and then slightly lower in the arthritic knee than in the control (Figure 1A). In the epiphyseal bone, the RBF was decreased in the arthritic knee on Day 1 of arthritis but then increased steadily and was significantly elevated on Days 7 and 14 (Figure 1B).

In the autoradiographs of 2-DG uptake the average densities calculated per area unit was 60–171% higher in the soft tissue of the arthritic knee than in the contralateral control (Figure 2). Due to the limited number of animals, no conclusion could be drawn on the differences in uptake between different days after challenge. The uptake was unevenly distributed, with marked accentuation in restricted areas adjacent to the cartilage of the femoral head and around the suprapatellar bursa. For the epiphyseal bone the average density/area unit was 12–18% higher in the arthritic joint.

When the total uptake of 2-DG was determined by scintillation technique, the levels were elevated about 100–150% on all days, with a slight peak on Day 3. These results are thus in accordance with those obtained by autoradiography. For [^{3}H]-thymidine, there was a slight increase in uptake on the first day of arthritis and a marked elevation on Day 3, but the levels did not significantly differ from the control knee on Days 7 and 14 (Figure 3). There was only a weak correlation between thymidine and glucose uptake (0.49, $p = 0.01$ in arthritic knee; 0.47, $p = 0.015$ in the control). A disturbing factor that may have influenced our results was the plasma levels of 2-DG versus [^{3}H]-thymidine: compared to the 5 min levels, between 30 and 40% of [^{3}H]-thymidine and 40–50% of 2-DG remained at 50 min. Since the relative water content, and hence possibly the distribution volume, was increased in the periarticular soft tissue of the arthritic knee, this could explain part of the increase in tissue levels. However, since the blood concentration (dpm/mg) of both [^{3}H]-thymidine and 2-DG was usually lower than the tissue concentration in the small intestine and the arthritic knee, we believe that the contribution of the circulating tracer levels to the results is minor. For instance, on Day 3 the mean concentrations of 2-DG in the small intestine and in the arthritic knee were 43 ± 12%,

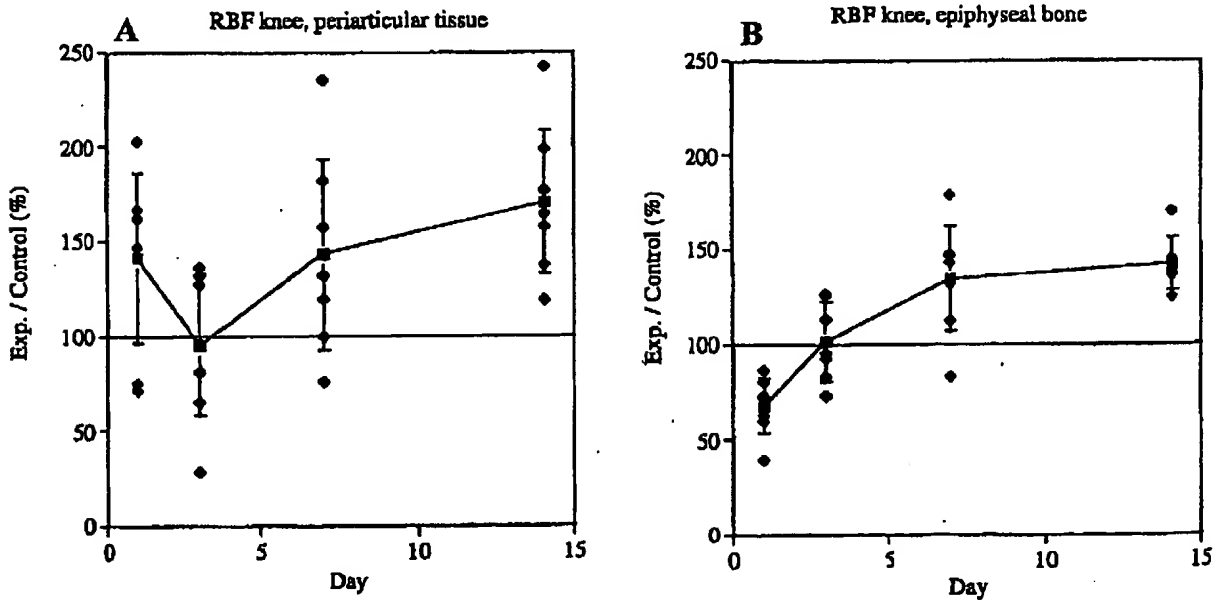

Figure 1. The weights related RBF (arthritis/control, %) in periarticular soft tissue (1A) and epiphyseal bone (1B). Results are given both as individual values and as means (95% confidence intervals).

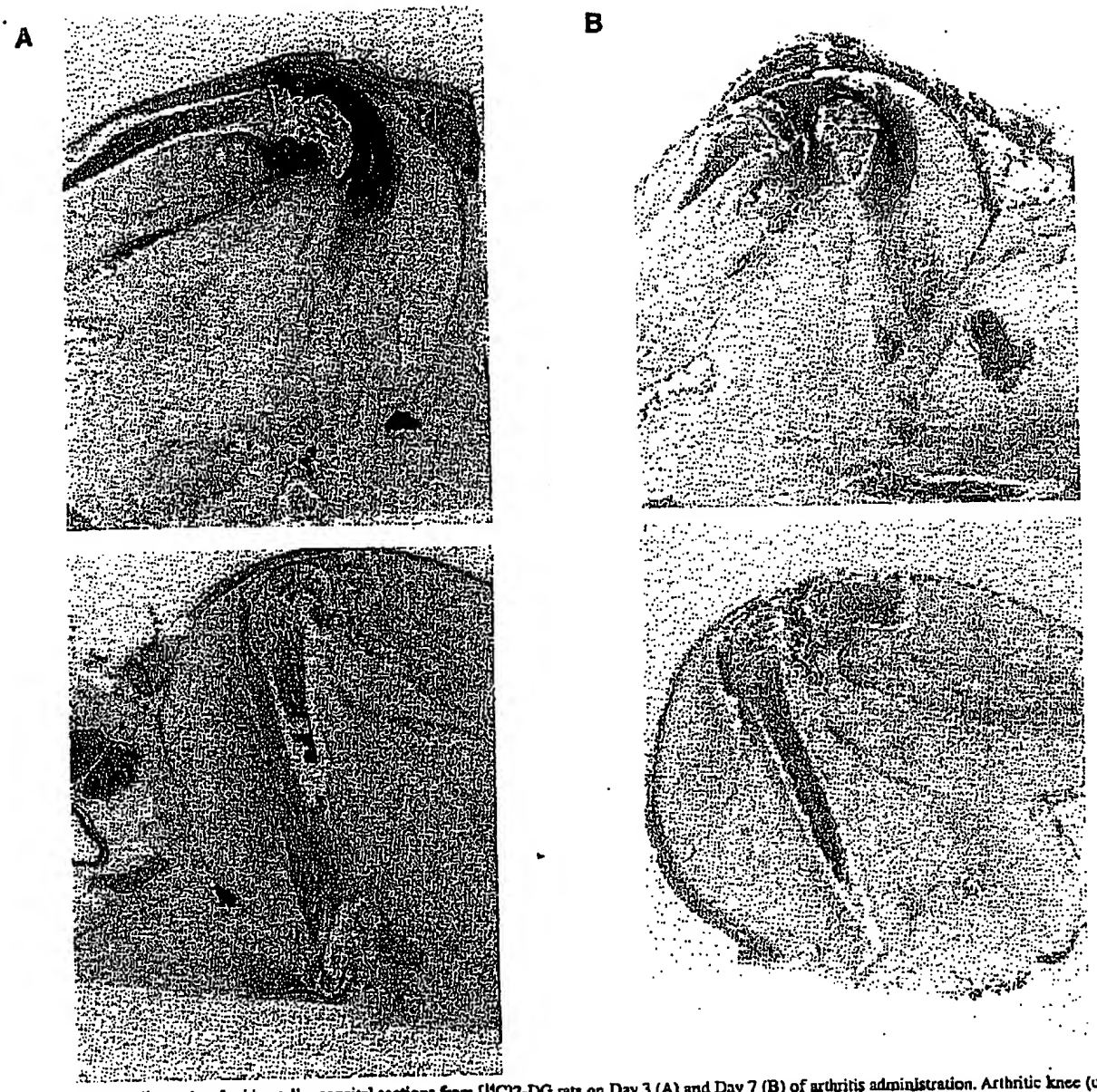

Figure 2. Autoradiographs of mid-patellar saggital sections from [^{14}C]2-DG rats on Day 3 (A) and Day 7 (B) of arthritis administration. Arthritic knee (upper panels) and contralateral control (lower panels). Note intense uptake in areas close to cartilage and around the suprapatellar bursa, which is distended on Day 3 (upper panel A).

61 ± 9% higher than in the blood, respectively. For [^{3}H]-thymidine the corresponding values were 844 ± 94% and 332 ± 12%, respectively.

When the ratio for weight related [^{3}H]-thymidine incorporation was plotted against the corresponding values for the pannus dry weight obtained in our adjacent paper[18], the observed increase was parallel between Days 1 and 3 but then differed markedly on the last 2 days of study (Figure 4). Both 2-DG uptake and [^{3}H]-thymidine incorporation were higher in the small intestine than in the control knee. In the arthritic joint, however, the 2-DG uptake was at about the same level as in the gut, whereas the [^{3}H]-thymidine incorporation was markedly lower. For example, on Day 3 of arthritis the values for 2-DG and [^{3}H]-thymidine content were 308 ± 6 and 40 ± 3 dpm/mg in the pannus tissue, respectively. Corresponding values for the small intestine were 278 ± 33 and 98 ± 14 and for the periarticular soft tissue of the control knee 124 ± 10 and 18 ± 1, respectively. The ratio between 2-DG and [^{3}H]-thymidine content was calculated to give an estimate of the amount of glucose

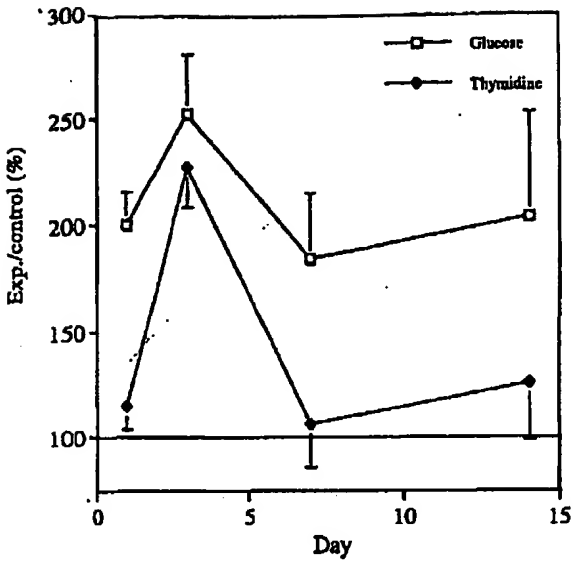

Figure 3. Weight related uptake (arthritis/control %; mean, with 95% confidence intervals) of [^{14}C]2-DG and [^{3}H]-thymidine in the periarticular soft tissue (n = 7 for Days 1, 3 and 7; Day 14 n = 5).

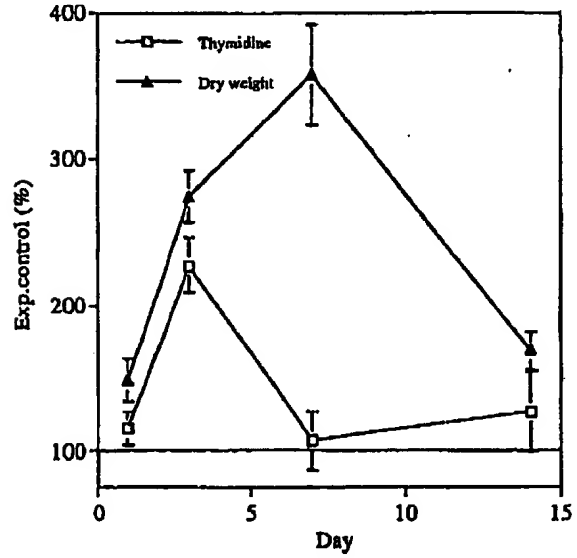

Figure 4. [^{3}H]-Thymidine uptake (dpm/mg) in the periarticular soft tissue compared with the increase in pannus dry weight from our companion paper[18] (both: arthritis/control %; mean, with 95% confidence intervals).

metabolism used for cell proliferation. This ratio was always lower in the small intestine than in the soft tissue of either knee. Comparing the knees, the ratio was significantly elevated in the arthritic knee on Days 1 and 7 of arthritis (Table 1).

As in our companion paper[18], indomethacin administered at Day 2 of arthritis reduced the vascular inflammation and pannus growth measured the next day, whereas the effect of

Table 1. Ratio for [^{14}C] 2-DG and [^{3}H]-thymidine content ([^{14}C]/[^{3}H]; both dpm/mg) in periarticular soft tissue of arthritic and control knees and small intestine on different days after arthritis induction. The ratio for the small intestine was the lowest in all experiments.

Day	Arthritic	Control (p)	Small Intestine
1	16.1 ± 2.7	9.3 ± 1.7 (0.013)	3.4 ± 0.3
3	8.0 ± 1.0	7.2 ± 1.0 (NS)	3.1 ± 0.4
7	15.2 ± 0.9	8.8 ± 0.9 (< 0.0001)	3.9 ± 0.5
14	17.3 ± 4.9	14.0 ± 6.8 (NS)	4.5 ± 1.3

Statistical significance was calculated between arthritic and control tissue using Student's 2 tailed t test for paired data. Periarticular tissue: n = 7 for Days 1–7; n = 5 for Day 14. Small intestine: n = 4–5.

MTX was smaller. Similar results were found in the present study: the difference in joint soft tissue weight between the knees was 232 ± 22 mg in the saline treated control group (n = 7), 172 ± 25 in the MTX group (n = 6, NS), and 156 ± 17 (n = 7) ($p < 0.05$ compared to control) in the indomethacin group. However, neither of the compounds affected the 2-DG uptake nor the [^{3}H]-thymidine incorporation. The ratios for the weight related uptake for 2-DG were: 308 ± 6%, 299 ± 12%, and 330 ± 14% for the control, MTX, and indomethacin groups, respectively. For [^{3}H]-thymidine, the corresponding values were 242 ± 37%, 224 ± 20%, and 242 ± 28%, respectively. These values are in good agreement with those found in the former series (Figure 4).

DISCUSSION

Blood flow; relation to intensity of vascular inflammatory response. The clinical response in AIA is a marked swelling of the arthritic knee, which peaks around Day 7 after challenge and is prominent for many weeks. Two variables may contribute to this measure: pannus growth and increased vascular porosity. In our companion paper[18], we found that the vascular inflammatory response, measured as leakiness for albumin, preceded the pannus growth and peaked around Day 3. In the present study, the RBF in the perivascular soft tissue of the arthritic knee was significantly elevated, compared to the control, only on Day 14. The time course for the changes was interesting: blood flow tended to increase on Day 1, drop to the lowest value on Day 3, and continued to increase thereafter in an inverse relationship with changes in vascular porosity. Hence, our findings suggest that, in this model, high vascular leakiness induces increased intraarticular pressure which, in turn, compresses synovial blood vessels; in other words, vascular inflammation negatively affects joint blood flow.

Glucose metabolism; localization and relation to blood flow. In contrast to RBF, 2-DG uptake was elevated more than 100% on all days studied. This discrepancy was especially marked on Day 3 when the mean RBF ratio was lowest and the 2-DG uptake was highest. This is compatible with the hypothesis that at that time, the oxygen supply would

not match glucose consumption and hence possibly lead to an ischemic situation. Conversely, ischemia may explain part of the increased 2-DG uptake since the energy yield per unit of glucose is much lower in anaerobic than in aerobic glycolysis. However, the present results do not allow conclusions regarding total energy metabolism, since we have no data on the consumption of triglycerides.

In addition to anaerobic glycolysis, it is possible that the 2-DG uptake is a marker of inflammatory activity. The autoradiographs indicated uneven distribution of 2-DG uptake, with "hot spots" in restricted areas close to the cartilage and around the suprapatellar bursa. Cationic antigens such as mBSA are deposited in the hyaline cartilage[24] and references) and it seems reasonable that the main foci of the inflammation lies adjacent to this antigen reservoir. In addition to the intense 2-DG uptake, autoradiographs revealed that the suprapatellar bursa was distended, and in a preliminary study using magnetic resonance imaging, marked plasma leakage seemed to occur in this area[25]. In our companion report[18] we also found a marked enhancement of cellular density in this area, which may explain the inflammatory activity and the augmented glucose uptake.

Relation between cellular proliferation rate and pannus growth. Local proliferation of fibroblast-like synovial cells has been observed in human rheumatoid synovia, but there is controversy about whether hyperplasia seen in the human disease is mainly caused by local proliferation or by influx of inflammatory cells[26]. In the present study the ratios (arthritis/control) for [^{3}H]-thymidine incorporation increased in tandem with the corresponding values for the dry weight of the soft tissue on the first and third days of arthritis, which proved to be the most intense phase of growth, before returning to baseline levels. This suggests that a substantial part of the initial pannus formation was due to cell proliferation, but that after a few days the increase in pannus weight may be more dependent on cellular influx or to increases in cell size. In our companion paper, we found that the pannus formation followed the inflammation in time and that it was probably induced by the latter. Since hypoxia is reported to enhance fibroblast proliferation[11] and induce release of growth factors such as vascular endothelial growth factor[27], it could be speculated that the presence of ischemia in the arthritic joint is an important link between inflammation and pannus formation.

Relation between cellular proliferation rate and glucose consumption. Effects of indomethacin and methotrexate. Cellular proliferation is an energy requiring process that, in addition to inflammatory activation and possible anaerobic glycolysis, may influence 2-DG uptake. We sought to obtain, by simultaneous co-administration of 2-DG and [^{3}H]-thymidine, a relative estimate of the amount of glucose used for cell proliferation. Among the compared tissues (the small intestine and the periarticular tissue in arthritic and control knees), the ratio between 2-DG and [^{3}H]-thymidine uptake was lowest, and the level of [^{3}H]-thymidine incorporation highest in the intestine, which is compatible with a high level of cell proliferation and a relatively large portion of the glucose being utilized for energy requiring processes associated with this. In the control knee the cell proliferation was low and the higher [^{14}C]/[^{3}H] ratio indicates that relatively more of the glucose was used in other processes such as basal metabolism. In the arthritic knee the 2-DG uptake was higher than in the control. A number of factors indicate that the extra glucose was not mainly used for cell proliferation. First, as mentioned, there was no marked enhancement of [^{3}H]-thymidine incorporation except on Day 3 and the [^{14}C]/[^{3}H] ratio was accordingly elevated compared to the control knee on Days 1 and 7. Second, on Day 3, when there was a peak in [^{3}H]-thymidine incorporation, the corresponding change in glucose metabolism was smaller and hence the [^{14}C]/[^{3}H] ratio returned to normal. Thus a marked increase in thymidine incorporation only affects the glucose metabolism to a minor extent. Third, there was only a weak correlation between 2-DG and [^{3}H]-thymidine levels in the periarticular soft tissue. This finding is in agreement with *in vitro* studies in which interleukin 1ß enhances glucose uptake by synoviocytes without enhancing cell proliferation[28]. The expression of this cytokine is elevated in murine AIA[29]; thus it seems likely that a large portion of the enhanced 2-DG uptake may be explained by the action of proinflammatory mediators that initiate energy requiring processes in the cells. However, there does not seem to be any clear coupling between glucose metabolism and vascular inflammation; on Day 14 there was still an increased metabolic activity in the joint measured both as 2-DG uptake and RBF, both in the soft tissue and the epiphyseal bone. This was not accompanied by significant inflammatory activity (measured as vascular leakiness) and the [^{3}H]-thymidine incorporation in soft tissue was not significantly elevated. In accordance, the reduction of vascular porosity by indomethacin on the third day of arthritis did not affect either glucose metabolism or cellular proliferation rate. Taken together, it seems that the glucose is mainly used for processes upstream of vascular inflammation but not directly correlated to this. It remains possible that some of the inflammation may be attributed to the influx of inflammatory cells into the arthritic joint, thereby changing the cell population to include cells with higher basal metabolic activity than the normal resident synoviocytes. This hypothesis is supported by the histological findings in our companion paper, where we found enhancements in cellular density close to the cartilage and, as stated above, around the suprapatellar bursa.

Methotrexate has, in the present dosage, antiinflammatory effects in the joint[18]. Our results with no marked effect on [^{3}H]-thymidine incorporation indicate that this dose of the drug does not markedly inhibit cell proliferation.

Blood flow and glucose metabolism in epiphyseal bone.

RBF in the epiphyseal bone was reduced in the arthritic knee on the first day of arthritis, then increased steadily, and was significantly elevated on Days 7 and 14, indicating that joint inflammation affects the bone. We have no obvious explanation for the initial decrease; it is possible that it represents a steal phenomenon. During the last 2 days of measurement the autoradiographic estimations of glucose uptake indicate that, in contrast to the findings in the soft tissue, the RBF is more elevated than the glucose uptake. This comparison thus supports the view that the blood flow in the soft tissue has a tendency to be insufficient.

In summary, our results indicate there is no increase in RBF during the first phase of arthritis despite a marked elevation in glucose metabolism. It remains possible that increased plasma extravasation has an attenuating effect on RBF. There is marked synovial hyperplasia characterized by an enhanced proliferation during the first days of arthritis. Most of the extra glucose metabolism is required for anaerobic glycolysis or processes other than cell proliferation, possibly related to the increased number of inflammatory cells. The main foci of the inflammation seem to be in the areas adjacent to cartilage.

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